MICROBIOLOGICAL WATER QUALITY IN LAUREL CREEK CONSERVATION AREA

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Ministry of the Environment

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MICROBIOLOGICAL WATER QUALITY IN LAUREL CREEK CONSERVATION AREA

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SUMMARY

A microbiological survey was conducted during June 19 to 23, 1978 to determine and evaluate the existing water quality in the Laurel Creek Conservation Area. The results indicated that microbiological water quality in the Laurel Creek Reservoir was generally good although low levels of fecal pollution existed. However, in the area upstream of the reservoir (the main inflowing streams) high bacterial densities indicated poor water quality. Taxonomic studies of fecal coliforms and fecal streptococci, isolated from both the reservoir and the inflowing streams, revealed that the fecal contamination was predominantly of non-human type and probably originated in surface runoff from nonpoint sources.

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1.0 INTRODUCTION

The surface waters of Ontario are subjected to many diverse uses, therefore, the management and preservation of good water quality is of great importance to the users. In particular, the maintenance of good water quality in recreational lakes and reservoirs is of vital concern since these areas are increasingly used for parks and cottage development, camp and picnic sites, swimming, bathing and other water recreational activities. The heavy usage of different bodies of water and surrounding land areas for recreational and other related activities exerts stress on the environment that may lead to water quality degradation.

Water quality deterioration predominantly occurs through microbiological contamination and/or nutrient enrichment (i.e. eutrophication), the latter stimulates and leads to undesirable and excessive growth of algae and other aquatic plants. Eutrophication generally appears slowly and lasts for considerably long periods, however microbiological contamination poses an immediate potential health hazard if the water is used for drinking and body contact recreational activities. A comprehensive account of health hazards associated with direct recreational contact with contaminated water is provided in a recent review on recreational water quality (1).

The major sources that cause nutrient enrichment include sewage and septic tank effluents, fertilizer losses, drainage and surface runoffs from urban/agricultural lands, forests, marshes and wetlands. Similarly, microbiological pollution originates from raw/inadequatly treated sewage, industrial effluents, and urban/rural runoffs from nonpoint sources.

In response to a request made by the Grand River Conservation Authority (GRCA), an intensive five-day study was conducted by the Ministry of the Environment in June 1978 to investigate the problem of water quality degradation in the Laurel Creek Conservation Area (LCCA). Previous work (2) indicated deteriorating microbial water quality in the LCCA to the extent that the swimming area was closed for public use in August 1977. In particular, coliform counts were consistently high both in the inflowing streams and the Laurel Creek Reservoir (Appendices III - VI).

The objectives of the present study were: (i) to examine and evaluate the existing microbiological water quality in the Laurel Creek Conservation Area, (ii) to ascertain the types and distribution patterns of fecal pollution indicator bacteria, and (iii) to determine the nature and sources of microbial contamination.

2.0 DESCRIPTION OF THE STUDY AREA

The main study areas were the Laurel Creek Reservoir (LCR), located northwest of the City of Waterloo in Waterloo township, and streams draining to the reservoir (Figure 1). The LCR has an area of 91.1 hectares and a mean depth of 1.62 meters with a water retention time estimated (during June 1978) to be 170 to 180 days. The main inflow, a combination of two streams, enters the northwest part of the reservoir and the outflow (controlled by a dam) is situated on the northeast side. The reservoir was originally constructed for flood control and stream flow augmentation, however, it is also used for recreational activities such as swimming, boating and picnicking. Several homes are scattered in the areas upstream of the reservoir.

The relevant study areas are located within the Laurel Creek watershed which has an area of approximately 7381.5 hectares (Figure 1) and covers parts of Woolwich, Wellesley, Wilmot and Waterloo townships in Waterloo county. The main source of surface water in the waterhshed is Laurel Creek, which

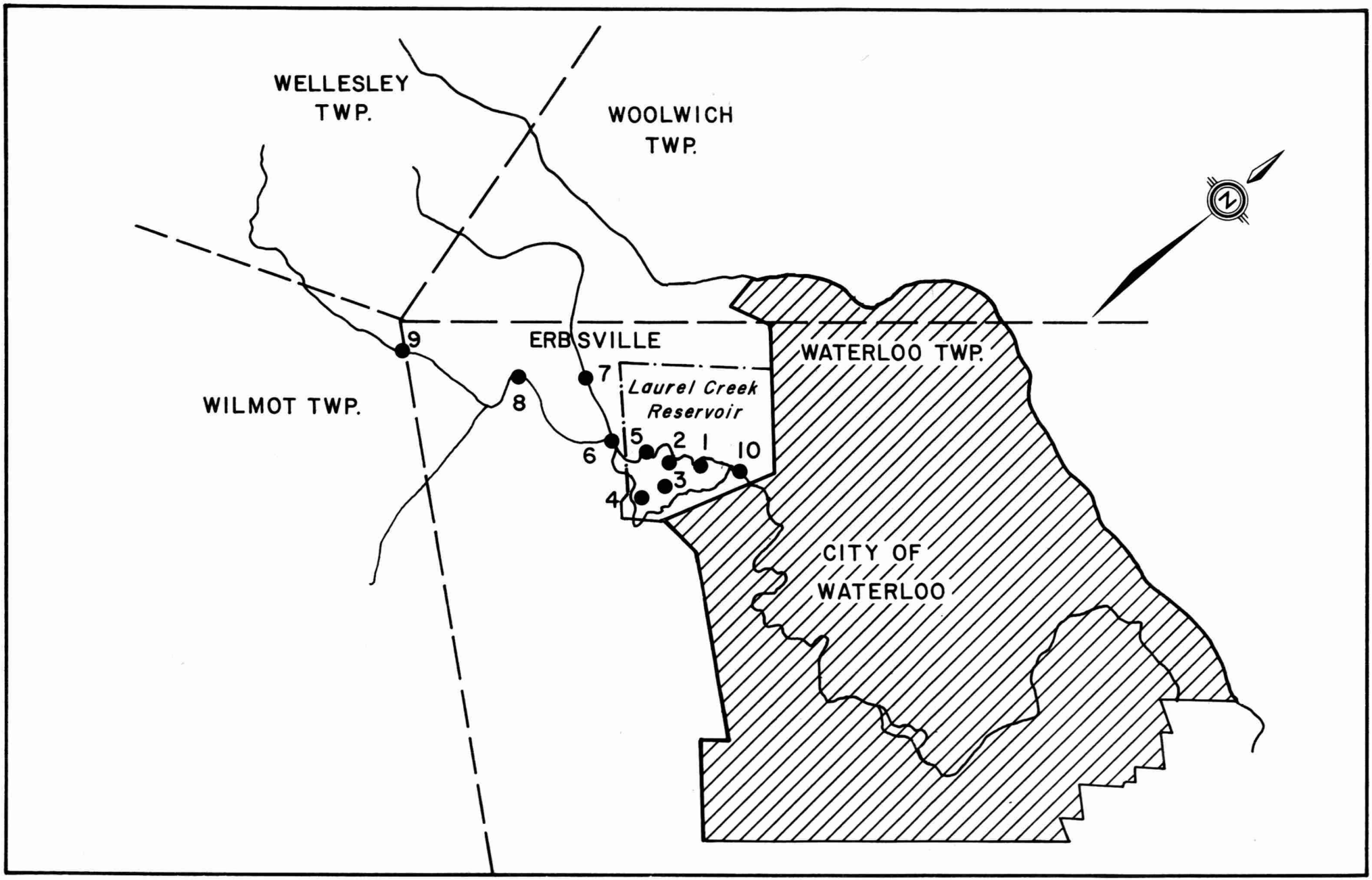

FIGURE 1 : LOCATION OF STUDY AREAS WITHIN LAUREL CREEK WATERSHED

drains into Grand River at the former town of Bridgeport, Ontario. The general land use for the entire Laurel Creek watershed (2) can be summarized as follows:

1) 28.2% Forest (forested land, woodlots, plantations, wetlands, secondary successions on old fields).
2) 51.1% Agriculture (cultivated crop fields, corn, mixed grain, managed hay and others).
3) 16.9% Others (idle land, non improved pasture, residential estates, homesteads).
4) 1.3% Open water (marshes, reservoirs).
5) 2.5% Roads.

3.0 DESIGN OF THE STUDY

3.1 Sampling Sites and Frequency of Sampling

During June 19 to 23, 1978, water samples were collected daily from four sites located on the main inflowing streams, five sites in the reservoir and one site at the outflow near the dam (Figure 2). In addition, sediment samples were obtained from all the reservoir locations on the first day of the survey. Water samples from the reservoir were taken 7 to 10 meters from shoreline and 1 meter below the surface, whereas at other sites samples were taken midway between the surface and bottom waters. Water samples were collected aseptically in sterile 500 ml polycarbonate bottles and sediment samples were taken in sterile Nalgene wide mouth jars (ca. 125 ml). All samples were stored on ice during transportation to the laboratory and were analyzed within 24 hours of collection.

3.2 Microbiological Parameters and Procedures

All water and sediment samples were analyzed for total coliforms (TC), fecal coliforms (FC), fecal streptococci (FS), Pseudomonas aeruginosa (PsA) and heterotrophic bacteria (HB). In addition, water samples from the beach area (Station 1) in the reservoir were tested for the presence of Candida albicans.

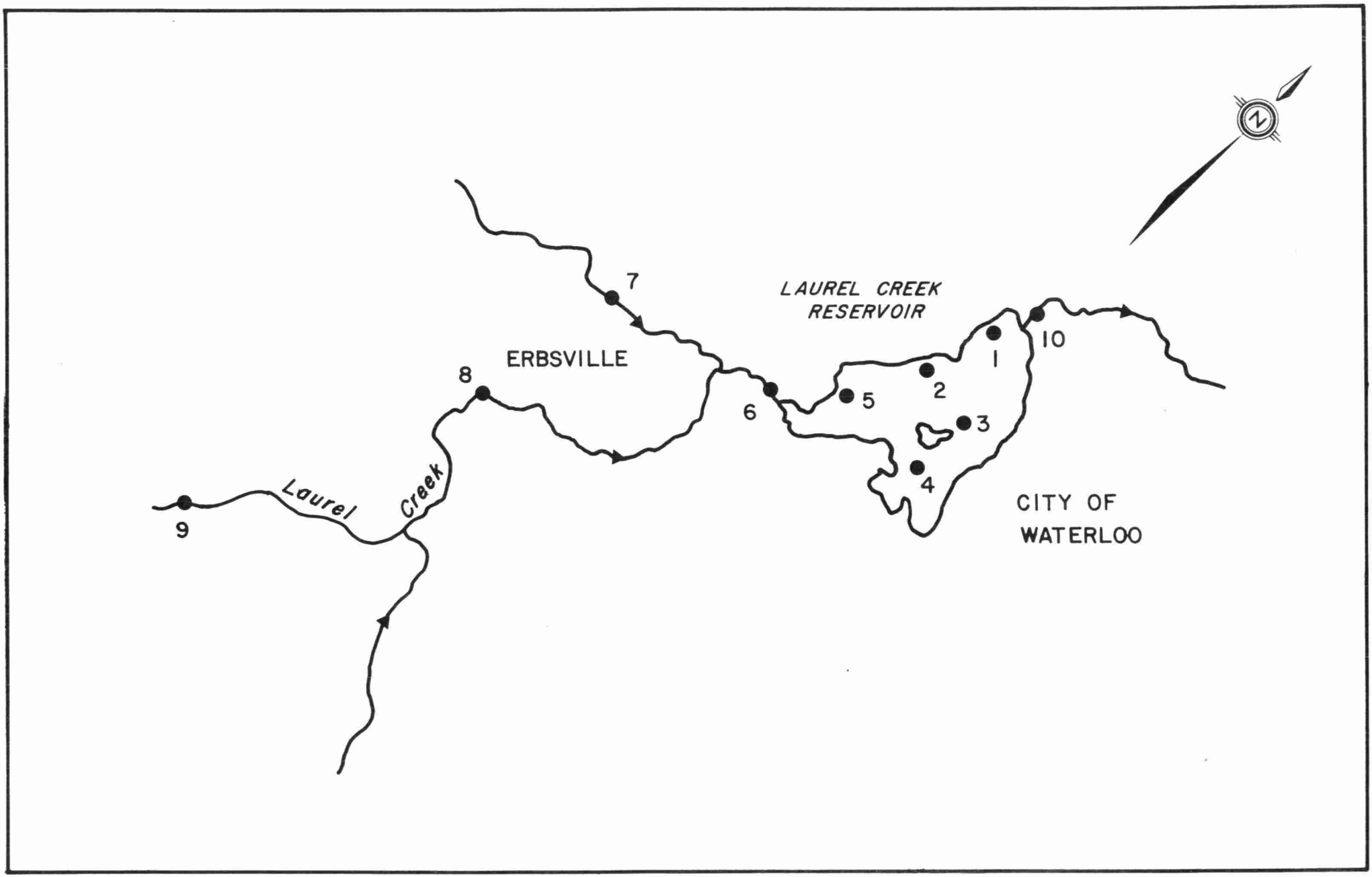

FIGURE 2 : LOCATION OF SAMPLING SITES IN THE LAUREL CREEK RESERVOIR, OUTFLOW AND INFLOWING STREAMS

The first three indicator organisms are all indigenous to man and other warm-blooded animals, and are generally found in large numbers in their feces. In addition, animal/human feces usually contain a variety of pathogens. Since many diseases common to man can be transmitted by feces, the probability of occurrence of these diseases is highest in areas where the water is contaminated with fecal material. Indicator organisms are used in water quality assessment to detect and indicate contamination from fecal material and hence the potential presence of pathogens.

Pseudomonas aeruginosa is recognized as a human opportunistic pathogen responsible for a diverse group of diseases including skin and upper respiratory infections and otitis externa, an outer ear infection (3). This organism is also found in feces and can be readily isolated from raw sewage samples. In recent years, P. aeruginosa has also been used as an indicator of fecal pollution (4).

Candida albicans (a yeast) constitutes a portion of the body's normal saprophytic microflora, but under appropriate conditions is an opportunistic pathogen causing a number of superficial skin infections. It is common in human/animal feces and raw sewage and has been found in both marine and fresh waters. Recently it has been suggested that C. albicans be considered as a potential indicator of water quality (5).

Heterotrophic bacteria are those bacteria which require organic carbon for their growth. The densities of these bacteria in water are influenced by the concentrations of organic nutrients and therefore may be a measure of the degree of organic enrichment in a given body of water.

The densities of total coliform (TC), fecal coliform (FC) and fecal streptococcus (FS) in water samples were determined using the membrane filtration (MF) technique. The methods used for their detection and enumeration are described in detail in the Ministry of the Environment's 'Handbook of Analytical Methods for Environmental Samples', Volume 2 (6). The levels of P. aeruginosa (PsA) were ascertained by the MF technique as described by Levin and Cabelli (3).

The MF procedure was employed for the detection of C. albicans as described by Buck and Bubucis (5). The levels of heterotrophic bacteria (HB) were obtained using the spot plate technique as described by Young (7). For all MF analyses, Gelman GN-6, 47 mm, 0.45 µm, white, gridded and autoclave-sterilized membrane filters were used.

The enumeration of pollution indicator bacteria in sediments was conducted by the most probable number (MPN) technique, following American Public Health Association Standard Methods (8). A 1/10 sediment dilution was prepared by adding 20 g (wet weight) sediment to 180 ml buffered water dilution blank and mixing in a Waring blender for one minute. From this suspension, further ten-fold dilutions were prepared using buffered water as the diluent.

TC densities were determined using a three dilution, five tube replication of lactose broth in a Standard MPN series. After incubation at 35°C for 48 h, inoculum from lactose broth tubes positive for gas was transferred into brilliant green lactose bile broth tubes and incubated at 35°C for an additional 48 h. All tubes showing gas production were recorded as confirmed TC for the purpose of the MPN computation.

For FC determinations, positive lactose broth tubes were used to inoculate EC broth tubes which were incubated at 44.5 $\pm$ 0.5°C for 24 h. Tubes showing growth and gas production were recorded as confirmed FC.

FS densities were estimated using three dilution, five tube replication of sodium azide dextrose broth series. Inoculated tubes were incubated at 35°C for 48 h. All tubes showing growth were transferred into tubes of ethyl violet azide broth which were then incubated at 35°C for an additional 48 h. Ethyl violet tubes showing turbidity and a purple pellet at the bottom of the tube were recorded as confirmed FS.

P. aeruginosa levels in sediments were enumerated by the MPN technique (8) using a three dilution, five tube series of modified Drake's medium 10 (9). The medium consisted of asparagine, 2.0 g; K_2HPO_4, 1.0 g; $MgSO_4.7H_2O$, 0.5 g;

K_2SO_4, 10.0 g; glycerol, 10.0 g per liter of distilled water. The tubes were incubated at 41.5 ± 0.5°C for 48-96 h, examined under long-wave ultraviolet light and those tubes showing greenish fluorescent pigment were interpreted as presumptive positive. All presumptive positive tubes were confirmed by streaking on modified Christensen's acetamide agar and skim milk agar. The acetamide agar contained acetamide, 10.0 g; NaCl, 5.0 g; K_2HPO_4, 1.4 g; KH_2PO_4, 0.7 g; $MgSO_4.7H_2O$, 1.54 g; phenol red, 0.012 g; agar, 15.0 g per liter of distilled water. The milk agar was prepared by dissolving 100.0 g skim milk (Difco) and 15.0 g agar into separate flasks each containing 500 ml distilled water. These two solutions were sterilized separately and were mixed prior to pouring into petri dishes.

Milk agar plates incubated at 35°C for 48-96 h, were observed for casein hydrolysis and production of a greenish fluorescent pigment. Acetamide agar plates were incubated at 41.5 ± 0.5°C for 48-96 h. Orange-red coloration around the colonies was considered as positive confirmation for P. aeruginosa MPN computation.

HB concentrations were determined in sediment using the same technique as outlined for water samples (7).

Fecal coliforms and fecal streptococci, obtained from MF analyses of waters, were respectively identified using Enterotubes (Roche Diagnostics, Hoffmann-LaRoche Inc., Nutley, New Jersey, U.S.A.) and procedures described by Pavlova et al (10) and Donnelly and Hartman (11).

3.3. Statistical Methods

The assessment of water quality cannot be determined accurately from a single water sample as microbial populations fluctuate tremendously in response to changing environmental conditions. Therefore, microbiological surveys require the collection of many samples from several stations over a designated period of time (3 to 5 days). The large amount of data generated is reduced by calculations to meaningful and easily managed statistics.

All microbiological data (355 determinations on 50 samples) collected during the five-day survey of the LCCA were transformed to logarithms (base 10), and all further analyses were done using the transformed data. The geometric mean (the most suitable central value) and variance were calculated for the values of TC, FC, FS, PsA and HB at every station providing concise data. The data were then analyzed by a one-way analysis of variance and Barlett's Test of Homogeneity to determine statistically significant variation in the microbial densities between stations or groups of stations. Using this procedure, the data from each station were tested against that of every other station until all stations with similar geometric means were separated into individual groups (e.g. Group A, B).

The group results and those for individual stations were identified by different stippling. Within each stippled area, the group geometric mean applied for each type of bacteria at all stations unless otherwise indicated by individual station values. In this manner, significantly different areas of the reservoir were differentiated as to the degree and level of microbial contamination.

4.0 RESULTS

4.1 Distribution and Levels of Bacteria in Water

The results of analysis of variance of all microbiological data and grouping of stations/areas are summarized in Appendices I and II.

In general, the waters in the Laurel Creek Reservoir (including the outflow) exhibited good quality as the bacterial densities (56 TC, 9 FC and 15 FS per 100 ml) were relatively low (Group A, Figure 3). However, the major inflowing streams showed degraded water quality as the concentrations of indicator bacteria (1600 TC, 130FC and 344 FS per 100 ml) were consistently higher (Group B, Figure 3) than those observed in the reservoir. An exception was station 9, where FC levels were less (35 FC per 100 ml) than those encountered at other locations downstream of this site.

A detailed bacteriological profile of the study area, showing geometric means of bacterial populations at all sites, is presented in Figure 4. The

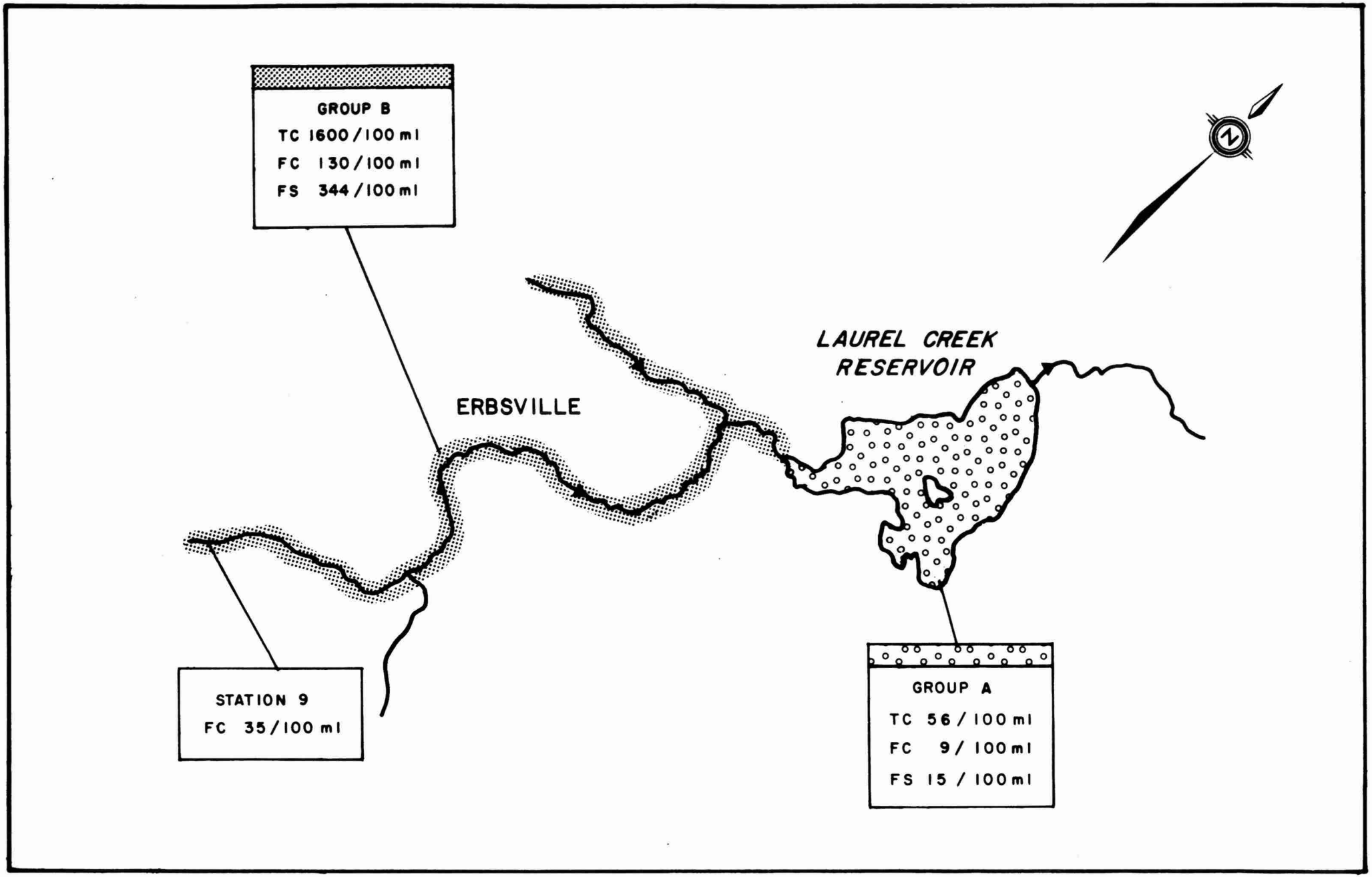

FIGURE 3 : DISTRIBUTION OF INDICATOR BACTERIA IN THE LAUREL CREEK CONSERVATION AREA (JUNE 19 TO 23)

concentrations of indicator bacteria were consistently high at the four upstream locations (Stations 6, 7, 8, 9). For example, the TC counts were always greater than 1000/100 ml, and FC and FS levels were generally greater than 100/100 ml. In addition, FC densities were always less than those of FS. As the water flowed down and entered the reservoir, however, a considerable reduction in bacterial populations occurred which may be attributed to the dilution effect, settling and/or bacterial die-off. In the reservoir and the outflow, TC, FC and FS densities were usually less than 100 organisms per 100 ml. Furthermore, with the exception of Stations 1 and 3, FS densities were higher than the concentrations of FC at the reservoir locations.

The results of taxonomic studies on 76 "fecal coliform" isolates, obtained from all sites monitored, indicated that all were fecal coliforms and most of them were E. coli (Table 1). Identification and differentiation of 82 "fecal streptococci", isolated from the study area, produced the results shown in Table 2. A majority (98%) of the isolates were confirmed as being fecal streptococci. Seven different species of the genus Streptococcus were isolated from stream locations, and four of these were also detected in the reservoir. Among the stream FS isolates, S. lactis, S. faecalis, S. faecium and S. avium were frequently isolated and accounted for 37.5, 18.8, 14.6 and 12.5 percent, respectively, of the total isolates. These four species were also found in the reservoir, where S. avium comprised 47 percent of the organisms isolated, followed by S. faecalis, S. lactis and S. faecium with 32.4, 11.8 and 8.8 percent, respectively.

The opportunistic pathogen P. aeruginosa was not isolated from any of the sites monitored with the exception of Station 5, where it was detected at a level of 32/100 ml only on the second day (June 20) of the survey. Likewise, C. albicans was not found in the bathing beach area (Station 1).

The heterotrophic bacterial densities in the study area had a similar pattern of distribution as that shown by the indicator bacteria. The concentrations of HB in the reservoir were 63,000/ml (Group A, Figure 5), but substantially higher

FIGURE 4 - BACTERIOLOGICAL PROFILE OF LAUREL CREEK RESERVOIR, OUTFLOW AND INFLOWING STREAMS SHOWING GEOMETRIC MEANS OF POPULATIONS OF TOTAL COLIFORMS (TC), FECAL COLIFORMS (FC) AND FECAL STREPTOCOCCI (FS)

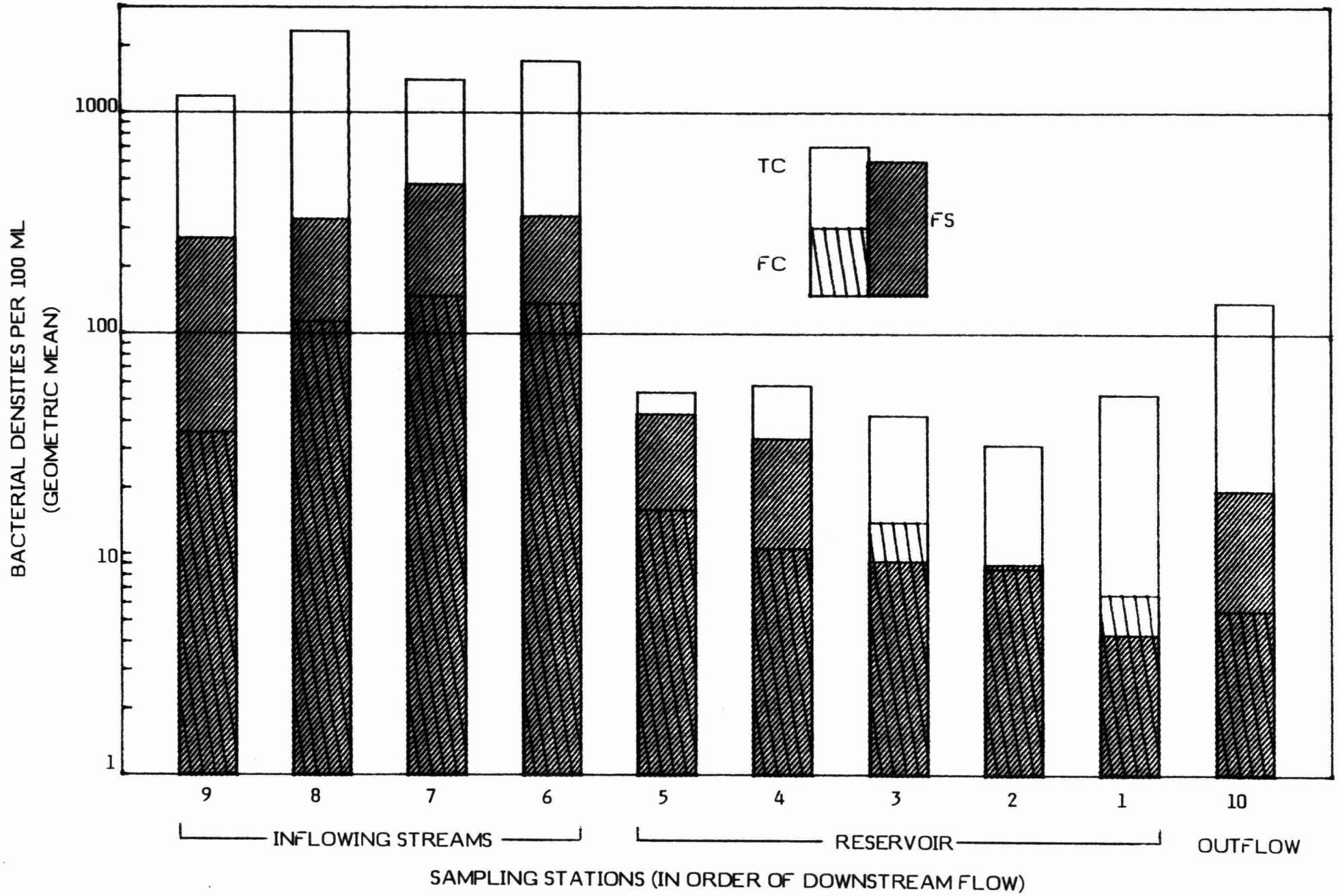

TABLE 1. IDENTIFICATION AND PERCENTAGE DISTRIBUTION OF FECAL COLIFORMS OBTAINED FROM DIFFERENT SITES IN THE LAUREL CREEK CONSERVATION AREA

Location	Organism	Percent Distribution
Inflowing streams (Stations 6, 7, 8, 9)	Escherichia coli	97.3 (36) *
	E. agglomerans	2.7 (1)
		(37) Total
Laurel Creek Reservoir (Stations 1, 2, 3, 4, 5 and outflow 10)	E. coli	100.0 (39)

* Values in parentheses are the number of confirmed isolates.

TABLE 2. IDENTIFICATION AND PERCENTAGE DISTRIBUTION OF FECAL STREPTOCOCCUS SPECIES ISOLATED FROM DIFFERENT SITES IN THE LAUREL CREEK CONSERVATION AREA

Location	Organism	Percent Distribution	
Inflowing streams (Stations 6, 7, 8, 9)	Streptococcus lactis	37.5	(18) *
	S. faecalis	18.7	(9)
	S. faecium	14.6	(7)
	S. avium	12.5	(6)
	S. cremoris	8.3	(4)
	S. faecalis var. liquefaciens	4.2	(2)
	S. faecium var. durans	2.1	(1)
	Non-fecal streptococcus	2.1	(1)
			(48) Total
Laurel Creek Reservoir (Stations 1, 2, 3, 4, 5 and outflow 10)	S. avium	47.1	(16)
	S. faecalis	32.3	(11)
	S. lactis	11.8	(4)
	S. faecium	8.8	(3)
			(34) Total

* Values in parentheses are the number of confirmed isolates.

densities (157,000/ml) were found in the inflowing stream locations (Group B, Figure 5) with the exception of Station 9 where the densities were 43,000/ ml.

4.2 Distribution and Levels of Bacteria in Sediment

Bacteriological data obtained from the analysis of sediments from the reservoir are given in Table 3. The densities of TC, FC, FS and PsA were very low, ranging from less than 1 to 5 per g wet weight. In contrast, the concentrations of HB were strikingly high (greater than 750,000 per g wet weight) at all sites.

4.3 Distribution and Concentrations of Chemical Parameters in Water

In addition to the microbiological analyses, water samples, collected from all sites on the first day of the survey, were analyzed for different chemical parameters.

The results of chemical analyses (conducted by the Water Quality Section using MOE's Standard Methods) are summarized in Table 4. The data indicated that the concentrations of all parameters in samples taken from the inflowing streams (Stations 6, 7, 8, 9) were considerably different from those in the reservoir (Stations 1, 2, 3, 4, 5). In particular, notable differences were detected in the levels of biochemical oxygen demand (BOD), total organic carbon (TOC), total Kjeldahl nitrogen (TKN), nitrate, total phosphorus (TP) and dissolved reactive phosphorus (DRP). In the outflow (Station 10), the concentrations of most of the parameters also differed appreciably from those observed at the other sites (particularly the inflowing streams) monitored.

5.0 DISCUSSION

5.1 MOE Study

During the study period, the water quality of the Laurel Creek Reservoir and its outflow was good as shown by low concentrations of indicator bacteria. The geometric mean densities of 56 TC and 9 FC per 100 ml were well

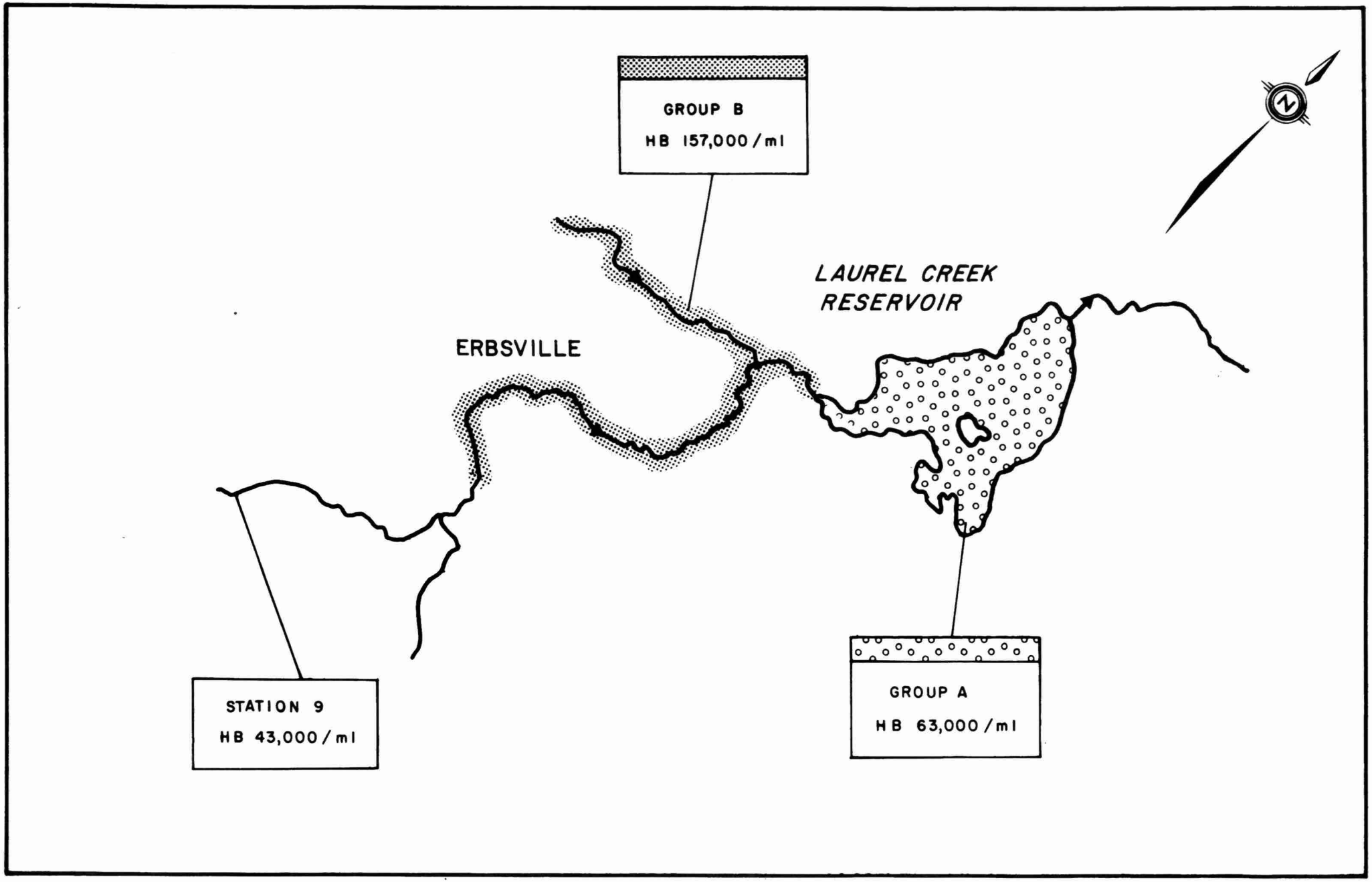

FIGURE 5 : DISTRIBUTION OF HETEROTROPHIC BACTERIA IN THE LAUREL CREEK CONSERVATION AREA (JUNE 19 TO 23)

TABLE 3. BACTERIAL POPULATIONS IN SEDIMENTS COLLECTED FROM THE LAUREL CREEK RESERVOIR SITES

STATION	BACTERIAL PARAMETER PER g WET WEIGHT				
	Total Coliform	Fecal Coliform	Fecal Streptococci	Pseudomonas aeruginosa	Heterotrophic Bacteria
1	2	< 1	< 1	< 1	> 750,000
2	2	< 1	< 1	< 1	> 750,000
3	2	1	< 1	< 1	> 750,000
4	< 1	< 1	< 1	< 1	> 750,000
5	5	< 1	< 1	< 1	> 750,000

TABLE 4. CONCENTRATIONS OF CHEMICAL PARAMETERS IN WATER SAMPLES COLLECTED ON JUNE 19 FROM DIFFERENT SITES IN THE LAUREL CREEK CONSERVATION AREA

STATION	CHEMICAL PARAMETERS (Milligrams Per Litre)										
	Biochemical Oxygen Demand	Chemical Oxygen Demand	CARBON			NITROGEN				PHOSPHORUS	
			Total	Total Organic	Inorganic	Free Ammonia	Total Kjeldahl	Nitrite	Nitrate	Total	Dissolved Reactive
INFLOWING STREAMS											
9	0.6	26	61.9	3.9	58.0	0.028	0.37	.011	1.34	0.028	0.019
8	1.4	10	62.3	4.3	58.0	0.008	0.52	.012	1.04	0.062	0.011
7	0.8	45	67.3	5.3	62.0	0.048	0.49	.028	0.820	0.045	0.029
6	1.0	84	63.2	4.2	59.0	0.020	0.46	.015	1.21	0.042	0.014
RESERVOIR											
5	4.0	65	60.1	9.1	51.0	0.006	0.94	.005	0.010	0.087	0.003
4	6.5	26	65.0	9.0	56.0	0.032	1.37	.007	0.040	0.122	0.003
3	4.0	41	60.4	9.4	51.0	0.036	1.30	.006	0.020	0.102	0.004
2	3.2	41	60.1	9.1	51.0	0.012	1.02	.010	0.010	0.093	0.003
1	5.5	65	60.1	9.0	52.0	0.020	1.31	.006	0.010	0.106	0.003
OUTFLOW											
10	4.0	39	61.0	9.0	52.0	0.014	1.25	.006	0.025	0.093	0.003

below the objectives of 1000 TC and 100 FC per 100 ml, recommended by the Ontario Ministry of the Environment for total body contact recreational activities (12). However, upstream of the reservoir, the microbiological water quality was generally poor as the geometric mean densities of coliforms exceeded the above mentioned objectives (Figure 4). Similarly, with the exception of Station 9, the levels of HB were higher in the inflowing streams than those in the reservoir (Figure 5) thereby further indicating degraded water quality and eutrophic conditions in these streams.

Among the opportunistic pathogens examined, C. albicans was not isolated from the beach area and P. aeruginosa was detected only once at Station 5. These results show that little health hazard existed in the reservoir during the course of this investigation.

In the sediments collected from the reservoir stations, the concentrations of indicator bacteria were lower than in the surface waters. This anomalous observation is difficult to explain since generally the number of both the indicators and pathogens are substantially higher in sediments than in the overlying waters (13). In contrast to low levels of indicator bacteria in sediments, the concentrations of HB were substantially higher in sediments than in the surface waters.

5.2 GRCA Study

During 1977 and 1978, GRCA conducted a series of water quality surveys of Laurel Creek Reservoir including its inflows and outflow. The results of these surveys partially described elsewhere (2) are summarized in Appendices III to VI. In general, these data indicated appreciable water quality degradation both in the reservoir and its major inflowing streams where levels of coliforms, particularly FC, were consistently high. For example, in the inflows (Appendices III and IV), the GM densities of FC were generally greater than 100 per 100 ml.

In most instances the GM concentrations of FC were higher than the corresponding TC densities at all sites monitored (Appendices III and IV). This is surprising because generally in polluted waters, FC (feces specific organisms) densities are lower than those of TC which are less feces specific in that they may originate from sources other than sewage and fecal matter. The membrane filter procedures used (Millipore Commercial Kit) for the analysis of total coliforms (on Endo medium) and fecal coliforms (on M-FC medium) may have been responsible for these peculiar results.

Although the GM densities of FC (Appendix V) in the beach area of the reservoir were less than 100 per 100 ml, individual values of greater than 100 were consistently observed in August and September during 1977 and 1978. Similarly, the GM levels of FC were higher than 100 per 100 ml during the September 1977 intensive survey (Appendix VI). These results indicated the existence of poor water quality in the reservoir particularly during the August-September period.

It is interesting to note that in the data presented in Appendices V and VI, the FC concentrations were lower than TC densities. This is probably because the samples, collected by GRCA during these surveys, were analyzed in the Ministry of Health (Kitchener, Ontario) Laboratory using APHA Standard Methods.

5.3 General Discussion

The results of the MOE investigation, in agreement with those of the GRCA study, indicated poor microbiological water quality in the inflows of the Laurel Creek Reservoir. The MOE data showed generally good microbiological water quality in the reservoir, however, the GRCA results indicated appreciable water quality degradation. The discrepancy in the results of these studies may be due to (i) differences in analytical procedures, (ii) more intensive GRCA surveys, (iii) seasonal differences and variation, and (iv) less human (recreational) use of the reservoir during the June (MOE) survey.

The bacteriological identification studies showed that the majority of the fecal coliform isolates from the inflows and the reservoir were E. coli. Identification of fecal streptococcus isolates revealed that four strains (viz. S. avium, S. lactis, S. faecalis and S. faecium) were common to both the inflows and reservoir. Although S. avium was the predominant (47%) strain in the reservoir, it accounted for only 13 percent of the inflowing stream isolates. The predominance of this particular species in the reservoir might have been due to significant bird populations inhabiting the beach and island areas within the reservoir. In addition, although about 38 percent of the stream isolates of streptococci were S. lactis, it was less prevalent (12%) in the reservoir.

The variety and types of streptococcus strains and generally high FS populations suggested that fecal pollution in the Laurel Creek Conservation Area was predominantly of non-human type and probably originated in overland runoff from non-point sources. The land use activities (i.e. agriculture related) in areas upstream of the reservoir as well as in the entire watershed support this conclusion. It appears that runoff transport fecal contamination from barnyards, feedlots, manured soil and from wild animals to the receiving surface waters.

Furthermore, the similarity in the types and distribution patterns of FC and FS suggested that the inflowing streams were the major source of existing fecal pollution in the Laurel Creek Reservoir. Although not apparent from the results of this study, a portion of the reservoir fecal contamination may have originated from sources within the reservoir. These sources include wildlife on the island and marshy areas, runoff from shoreline and heavy human/bird usage of the beach area (2).

The results of chemical analyses indicated that the levels of BOD, TOC, TKN and TP were higher in the reservoir than in the inflowing streams, however, the reverse was observed with respect to the nitrate levels. The assimilation of nitrate to organic nitrogen indicates substantial primary production

in the reservoir. Likewise, the increase in BOD, TOC, and TP levels in the reservoir indicates additional sources (other than streams, e.g. resuspension of sediments) of these components, but it is also consistent with increased primary productivity in the reservoir.

6.0 CONCLUSIONS

The results of this investigation showed good bacteriological water quality in the Laurel Creek Reservoir, however, water quality was generally poor in the main inflowing streams. In particular, the levels of fecal pollution indicators and heterotrophic bacteria were elevated at Stations 6 and 8, located on Laurel Creek downstream of Station 9, as well as at a nearby tributary site (Station 7).

Taxonomic examination of $\bar{F}C$ and $\bar{F}S$ isolates, obtained from all monitored sites, indicated that fecal pollution was mainly of non-human type originating in surface runoff from diffuse sources. Moreover, the major inflowing streams appear to be the main source of bacterial contamination in the Laurel Creek Reservoir.

In general, the microbiological water quality was not significantly influenced by the limited recreational use of the reservoir during the survey (June 1978) period. Nevertheless, it is reported (14) that in recreational waters the densities of indicator bacteria generally increase from extensive human use, especially during weekend periods. Therefore, it is probable that high bacterial input from the inflowing streams together with the input from increased human use (mostly experienced during July and August in Ontario) could result in substantial water quality deterioration and potential health hazards in Laurel Creek Reservoir.

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APPENDICES

EXPLANATION OF TERMS IN APPENDICES I - II

GM - the geometric mean of the bacterial level.

t - the calculated test of significance or student t-test used to compare stations, groups and a survey.

If t for the number of degrees of freedom shown is greater than the critical t value, a significant difference (SD) occurs.

Log GM - the logarithm (base 10) of the geometric mean.

S.E. - the standard error of the log GM where

$$S.E. = \frac{s}{\sqrt{N}}$$ and s = standard deviation

N - the number of values in the mean.

APPENDIX I

ANALYSIS OF VARIANCE SUMMARY FOR TOTAL COLIFORMS, FECAL COLIFORMS, AND FECAL STREPTOCOCCI (PER 100 ML)

GROUPS	Log GM	SE	N	GM
TOTAL COLIFORMS				
Group A (Stations 1, 2, 3, 4, 5, 10)	1.7447	0.0690	30	55.6
Group B	3.2015	0.0567	20	1590.5
FECAL COLIFORMS				
Group A (Stations 1, 2, 3, 4, 5, 10)	0.9736	0.0827	30	9.4
Group B (Stations 6, 7, 8)	2.1151	0.0532	15	130.3
Station 9	1.5481	0.1056	5	35.3
FECAL STREPTOCOCCI				
Group A (Stations 1, 2, 3, 4, 5, 10)	1.1643	0.1205	30	14.6
Group B (Stations 6, 7, 8, 9)	2.5360	0.0541	20	343.6

APPENDIX II

ANALYSIS OF VARIANCE SUMMARY FOR HETEROTROPHIC BACTERIA PER ML

GROUPS	Log GM	SE	N	GM
Group A (Stations 1, 2, 3, 4, 5, 10)	4.8013	0.0541	30	63288.8
Group B (Stations 6, 7, 8)	5.1950	0.0908	15	156658.5
Station 9	4.3304	0.1009	5	42577.9

EXPLANATION OF APPENDICES III - VI

Data summarized in Appendices III, IV, V and VI were collected during a series of water quality surveys of the Laurel Creek Conservation Area conducted in 1977 and 1978 by the Grand River Conservation Authority, Cambridge, Ontario.

APPENDIX III

GEOMETRIC MEAN (GM) AND RANGE (R) OF TOTAL COLIFORM AND FECAL COLIFORM POPULATIONS (PER 100 ML) IN INFLOWS AND OUTFLOW OF LAUREL CREEK RESERVOIR [1]

	TOTAL COLIFORM						FECAL COLIFORM					
	1977[2]			1978[2]			1977			1978		
STATION	GM	R	N[3]	GM	R	N	GM	R	N	GM	R	N
Inflowing Streams												
1	93.5	10 - 514	8	58.9	10 - 1100	9	80.4	10 - 320	10	130.6	16 - 640	13
2	84.5	10 - 360	8	163.1	20 - 1200	13	129.9	22 - 450	9	190.3	72 - 1000	13
3	85.8	6 - 300	10	154.2	40 - 800	12	131.5	14 - 500	10	129.0	52 - 570	13
4	74.8	28 - 210	8	116.6	20 - 406	13	127.6	14 - 500	10	178.1	90 - 1150	13
5	47.1	16 - 120	11	70.7	2 - 280	13	89.7	26 - 300	9	56.4	12 - 286	13
Outflow												
6	54.7	6 - 200	9	32.8	8 - 60	8	156.6	76 - 312	8	111.3	68 - 232	13

1 Data obtained during Grand River Conservation Authority's (GRCA) Weekly/Bimonthly Water Quality Surveys; samples were analyzed in GRCA Laboratory using Millipore's Commercial Kit for total coliform and fecal coliform analyses.

2 1977 (June - December), 1978 (June - September).

3 Number of samples from which acceptable data were available for calculating the R and GM Values.

APPENDIX IV

GEOMETRIC MEAN (GM) AND RANGE (R) OF TOTAL COLIFORM AND FECAL COLIFORM POPULATIONS (PER 100 ML) IN THE LAUREL CREEK RESERVOIR INCLUDING THE INFLOW AND OUTFLOW [1]

	TOTAL COLIFORM			FECAL COLIFORM		
STATION	GM	R	N[2]	GM	R	N
Inflow	128.6	6 - 400	15	286.7	24 - 800	18
Reservoir						
3	38.7	8 - 240	14	67.5	2 - 1000	19
2	10.9	2 - 62	11	45.7	2 - 700	16
1	21.4	4 - 60	12	40.4	4 - 306	15
Outflow	32.9	5 - 200	12	147.0	20 - 312	18

[1] Data obtained during May to September, 1977 as a part of GRCA's Weekly Water Quality Surveys; samples were analyzed in GRCA Laboratory using Millipore's Commercial Kit for total coliform and fecal coliform analyses.

[2] Number of samples from which acceptable data were available for calculating the R and GM Values.

APPENDIX V

GEOMETRIC MEAN (GM) AND RANGE (R) OF TOTAL COLIFORM AND FECAL COLIFORM POPULATIONS IN THE BEACH AREA OF LAUREL CREEK RESERVOIR [1]

STATION	TOTAL COLIFORM						FECAL COLIFORM					
	1977			1978			1977			1978		
	GM	R	N[2]	GM	R	N	GM	R	N	GM	R	N
1	123.7	2 - 1400	18	73.4	2 - 2400	15	44	2 - 700	18	69	4 - 2200	13
2	148.4	6 - 3600	18	82.2	2 - 8000	15	93.8	4 - 1200	17	59.8	2 - 2800	14
3	133.6	4 - 3200	18	73.4	10 - 800	11	58.9	2 - 700	18	30.6	2 - 300	11

[1] Data obtained during May to September, 1977 and 1978 as a part of GRCA's Weekly Water Quality Surveys; samples were analyzed in Public Health (Kitchener) Laboratory using APHA Standard Methods for total coliform and fecal coliform analyses.

[2] Number of samples from which acceptable data were available for calculating the R and GM Values.

APPENDIX VI

GEOMETRIC MEAN (GM) AND RANGE (R) OF TOTAL COLIFORM, FECAL COLIFORM AND FECAL STREPTOCOCUS POPULATIONS AT STATIONS LOCATED IN LAUREL CREEK AND THE RESERVOIR [1]

	TOTAL COLIFORM			FECAL COLIFORM			FECAL STEPTOCOCCUS		
STATION	GM	R	N[2]	GM	R	N	GM	R	N
Outflow	308.4	100 - 500	9	96.5	28 - 600	9	24.7	12 - 52	9
Reservoir									
1	548.6	200 - 1500	10	164.7	36 - 1100	10	64.1	4 - 200	10
2	546.8	100 - 2400	10	336.9	56 - 1400	10	73.7	14 - 300	10
3	563.7	200 - 2400	10	237.7	90 - 800	10	74.5	14 - 500	10
4	914.2	300 - 2400	10	420.4	96 - 1200	10	72.2	14 - 200	10
5	1088.1	400 - 2200	10	176.1	72 - 1000	10	37.3	4 - 500	10
Inflowing Streams									
6	537.2	100 - 2300	10	170.7	62 - 700	10	35.5	8 - 200	10
7	422.0	200 - 1000	10	108.4	34 - 300	10	40.4	2 - 400	10
8	314.5	100 - 1500	10	125.0	32 - 800	10	61.5	12 - 600	10
9	435.5	64 - 1900	10	110.3	30 - 200	10	48.3	10 - 300	10
10	682.9	60 - 2800	10	179.6	46 - 2800	10	159.1	20 - 800	10
11	957.1	200 - 4000	10	269.2	30 - 3000	10	81.9	4 - 1400	10
12	259.7	100 - 800	10	56.1	20 - 128	10	25.9	4 - 300	10
13	1256.5	300 - 4000	10	283.1	62 - 2100	10	44.1	12 - 200	10

[1] Data obtained during September 28 to October 17 (1977), as a part of GRCA's Intensive Water Quality Survey; samples were analyzed in Public Health (Kitchener) Laboratory using APHA Standard Methods for total coliform, fecal coliform and fecal streptococcus analyses.

[2] Number of samples from which acceptable data were available for calculating the R and GM Values.

	DATE DUE		